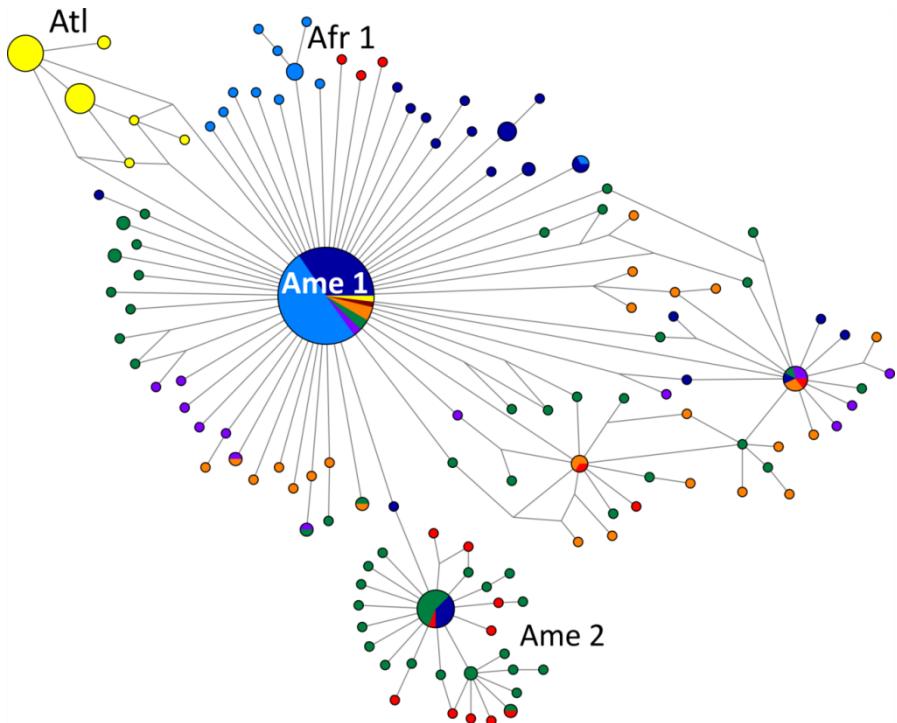


Supplementary Figures, Tables, and Text

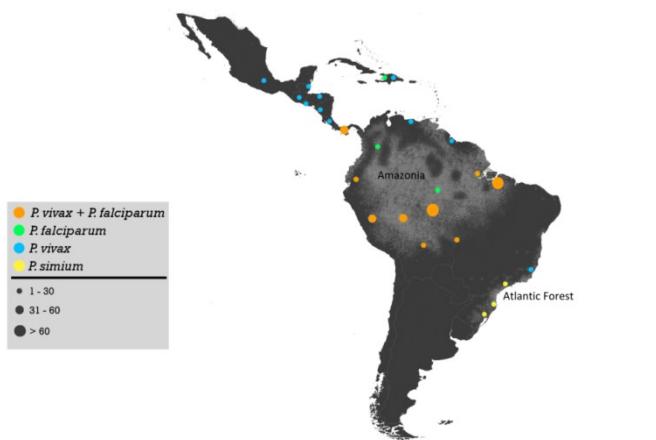
Human migration and the spread of malaria parasites to the New World

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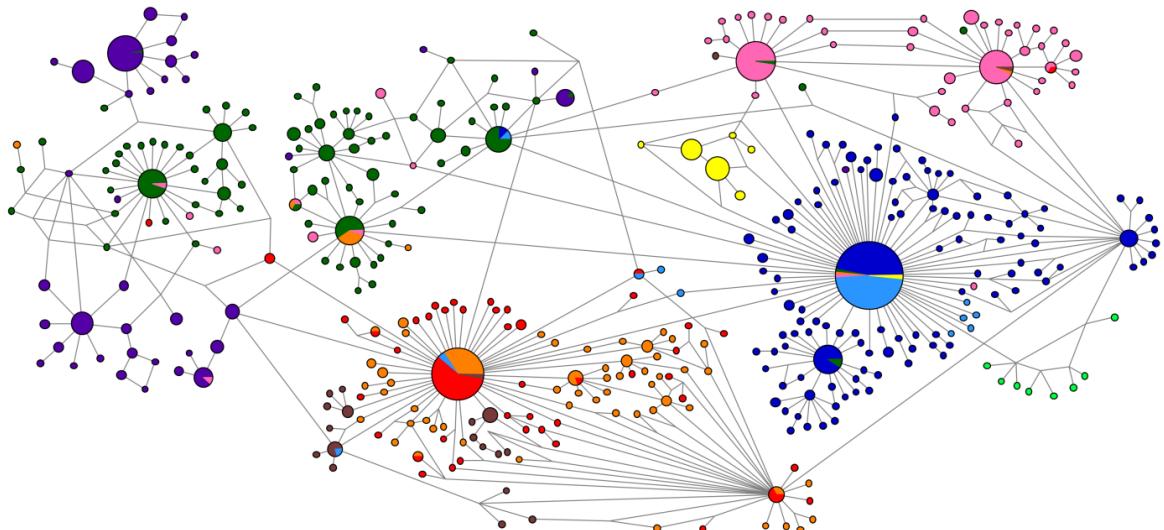
Supplementary Figures



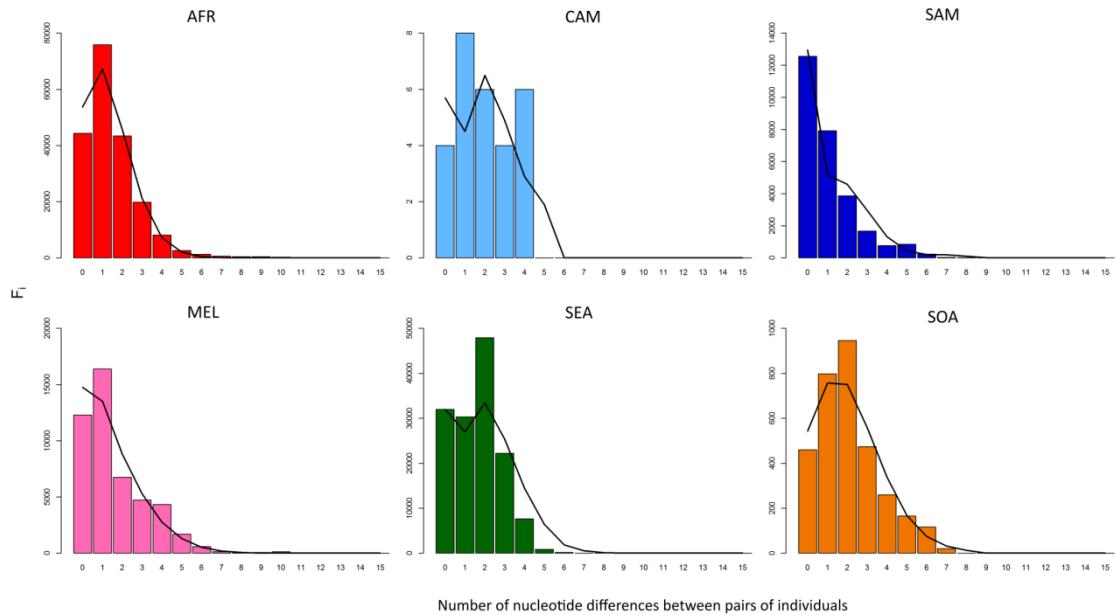
Supplementary Fig. 1. Median-joining network of *Plasmodium vivax*/P. *simium* mitochondrial lineages from South and Central America. Circle sizes are proportional to haplotype frequencies, and pairs of haplotypes connected by a straight line differ by a single mutational step. Clades Ame 1, Ame2, Atl, and Afr1 are identified in the network. The following color code was used to indicate the geographic origin of isolates: dark blue = Brazil; light blue = Central America and Mexico; orange = Peru; green = Colombia; dark red (wine) = Ecuador; yellow = Atlantic Forest of Southeast and South Brazil, and red = Venezuela.



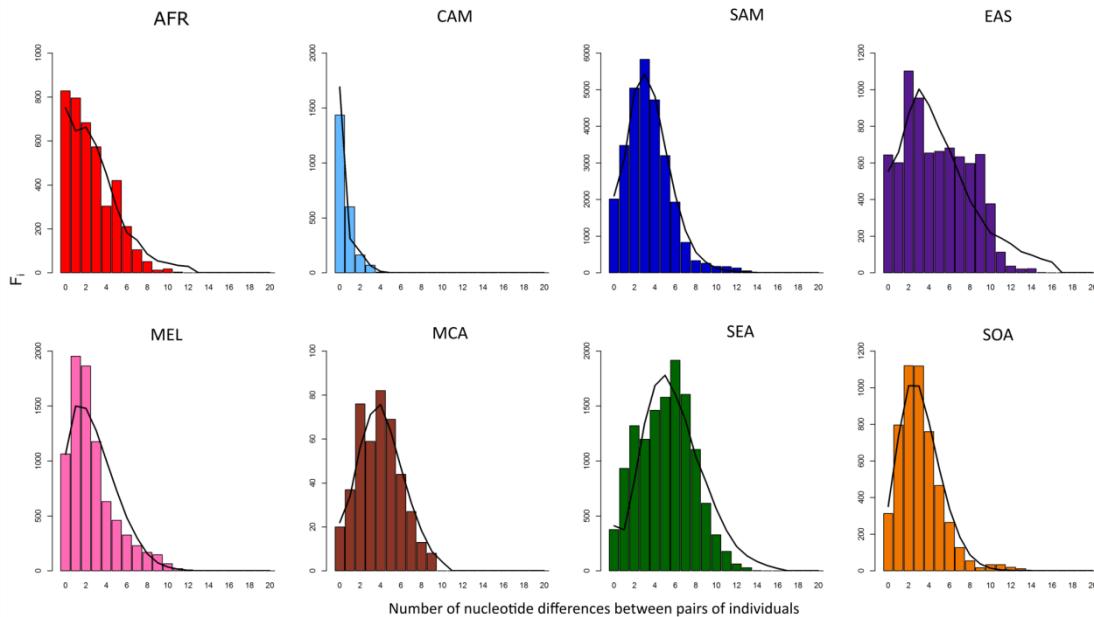
Supplementary Fig. 2. Map showing the collection sites of the New World samples of *Plasmodium falciparum* and *P. vivax/P. simium* analyzed in this study. The map was built using the open-access R software library *rworldmap: mapping global data* combined with the *ggplot2* library, which are both available at <http://www.R-project.org/>.



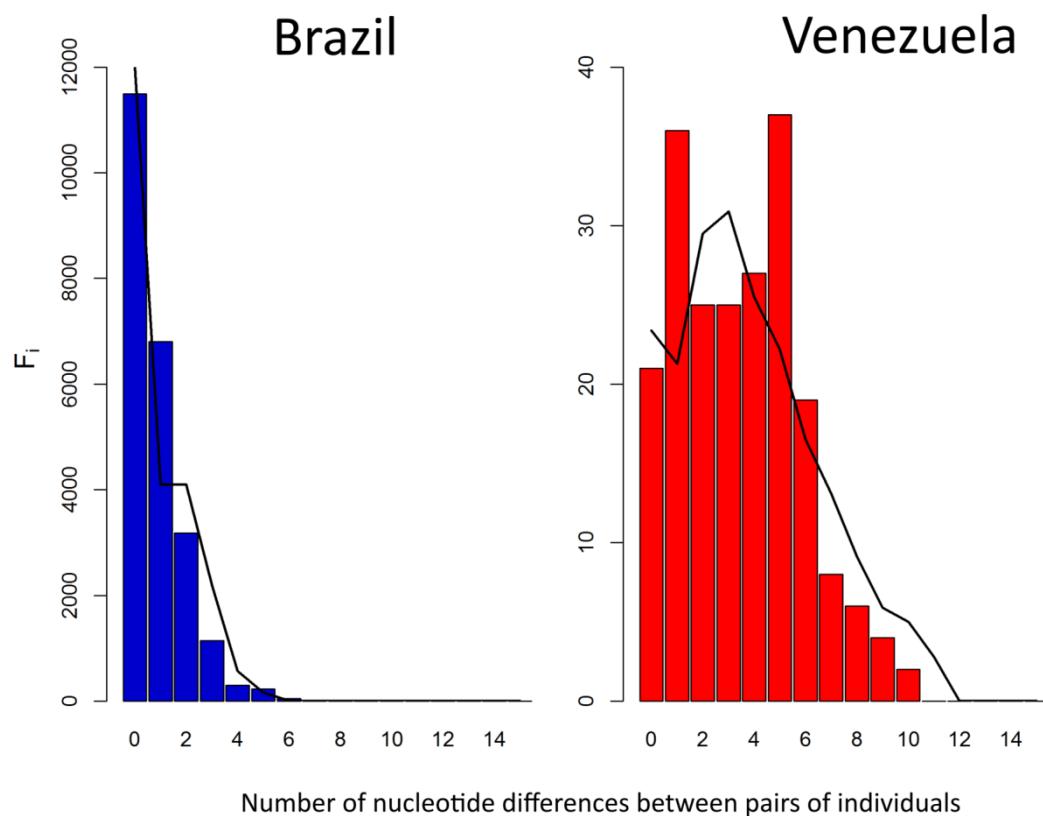
Supplementary Fig. 3. Median-joining network of the global sample of *Plasmodium vivax* mitochondrial lineages, including *P. vivax*-like parasites from African great apes. As in Fig. 2b, circle sizes are proportional to haplotype frequencies, and pairs of haplotypes connected by a straight line differ by a single mutational step. The following color code was used to identify the geographic origin of parasites: red = Africa (human samples), light green = African great apes (data from²⁷), dark blue = South America, light blue = Central America and Mexico, yellow = Atlantic Forest from southeast and South Brazil, brown = Middle East and Central Asia, orange = South Asia, green = Southeast Asia, dark purple = East Asia, and pink = Melanesia.



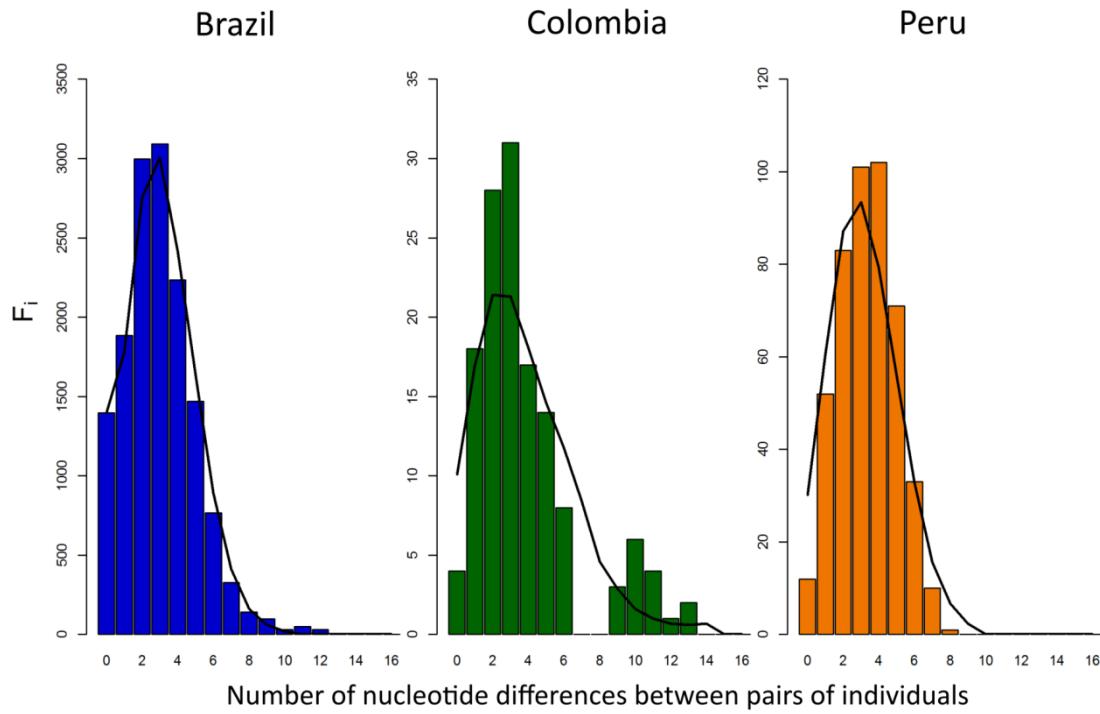
Supplementary Fig. 4 Mismatch distribution analysis of mitochondrial lineages from regional *Plasmodium falciparum* populations. The following populations were analyzed: Africa (AFR; red), South America (SAM; dark blue), Central America (CAM; light blue), South Asia (SOA; orange), Southeast Asia (SEA; green), and Melanesia (MEL; pink). Bars show observed frequencies, and continuous lines show expected frequencies under a sudden population expansion model⁷⁶. Note that observed and expected distributions are quite similar in most populations, except for CAM ($n = 8$ isolates) (see also S11 for significance tests).



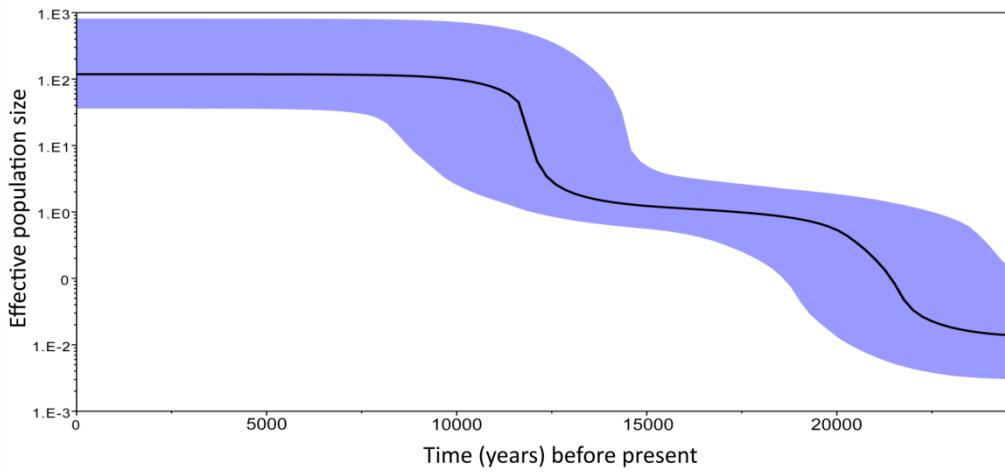
Supplementary Fig. 5. Mismatch distribution analysis of mitochondrial lineages from regional *Plasmodium vivax* populations. The following populations were analyzed: Africa (AFR; red), South America (SAM; dark blue), Central America and Mexico (CAM; light blue), Middle East and Central Asia combined (MCA; brown), South Asia (SOA; orange), Southeast Asia (SEA; green), East Asia (EAS; dark purple), and Melanesia (MEL; pink). Bars show observed frequencies, and continuous lines show expected frequencies under a sudden population expansion model⁷⁶. Note that observed and expected distributions are quite similar in all populations (see also Supplementary Table 11 for significance tests).



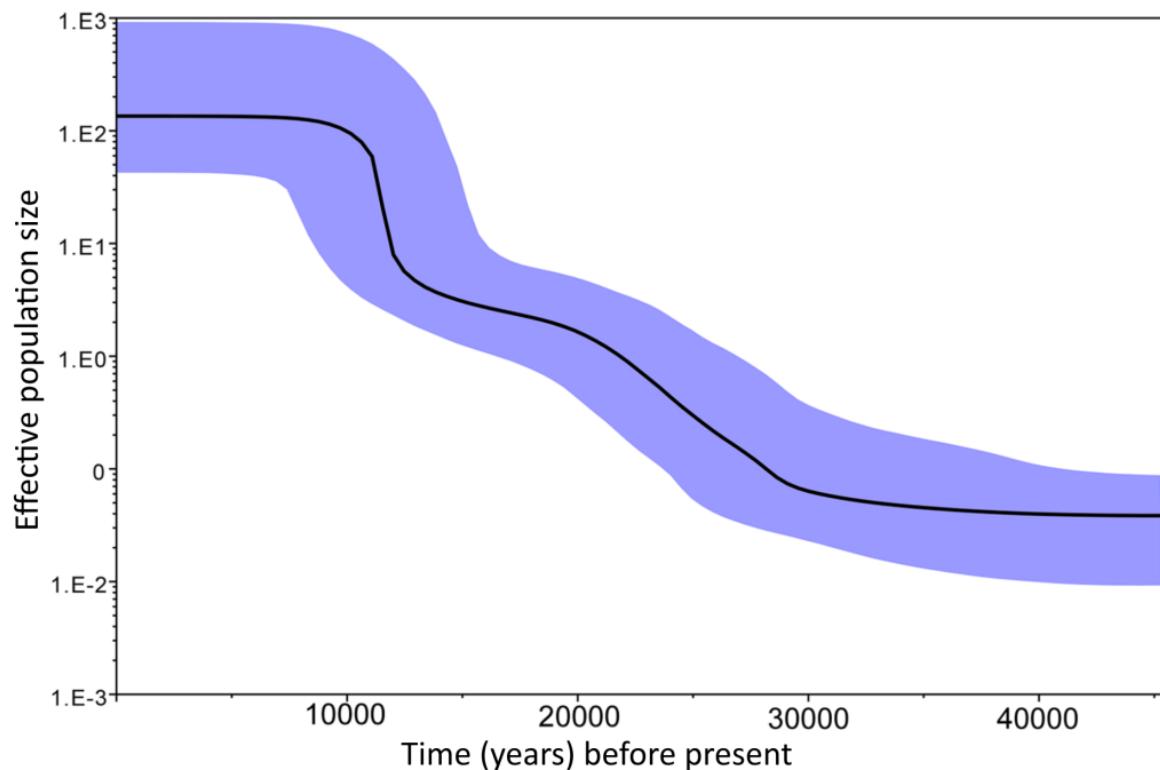
Supplementary Fig. 6. Country-specific mismatch distribution analyses of mitochondrial lineages from *Plasmodium falciparum* populations from South America. Bars show observed frequencies, and continuous lines show expected frequencies under a sudden population expansion model⁷⁶. See Supplementary Table 12 for significance tests.



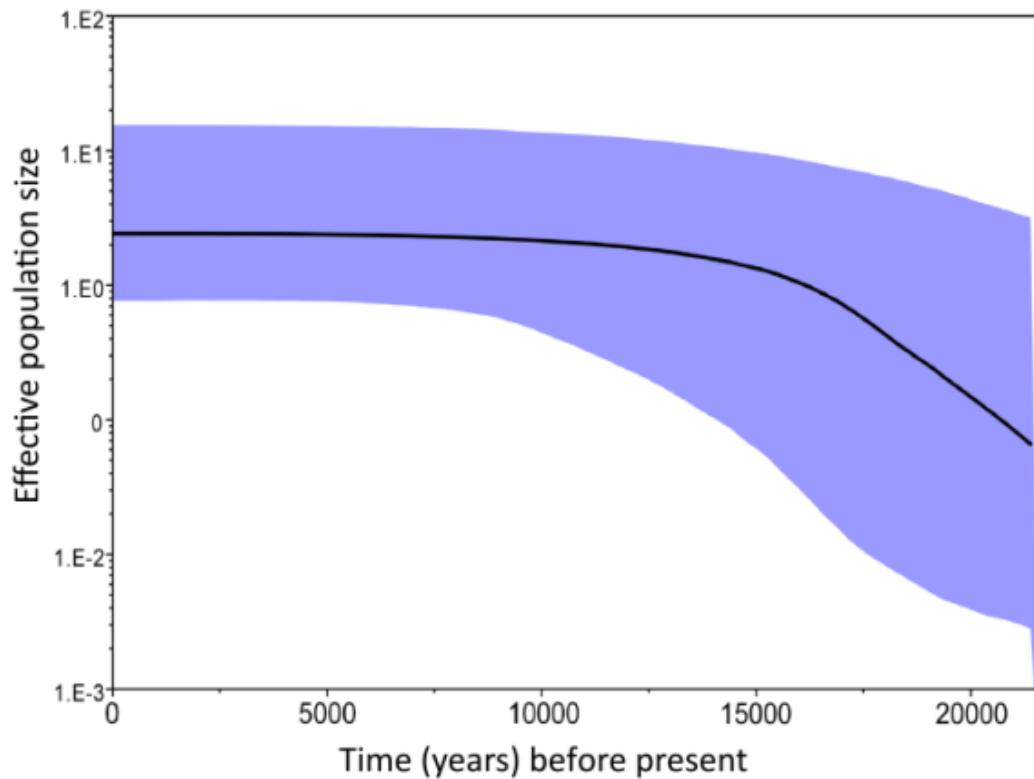
Supplementary Fig. 7. Country-specific mismatch distribution analyses of mitochondrial lineages from *Plasmodium vivax* populations from South America. Bars show observed frequencies, and continuous lines show expected frequencies under a sudden population expansion model⁷⁶. See Supplementary Table 12 for significance tests.



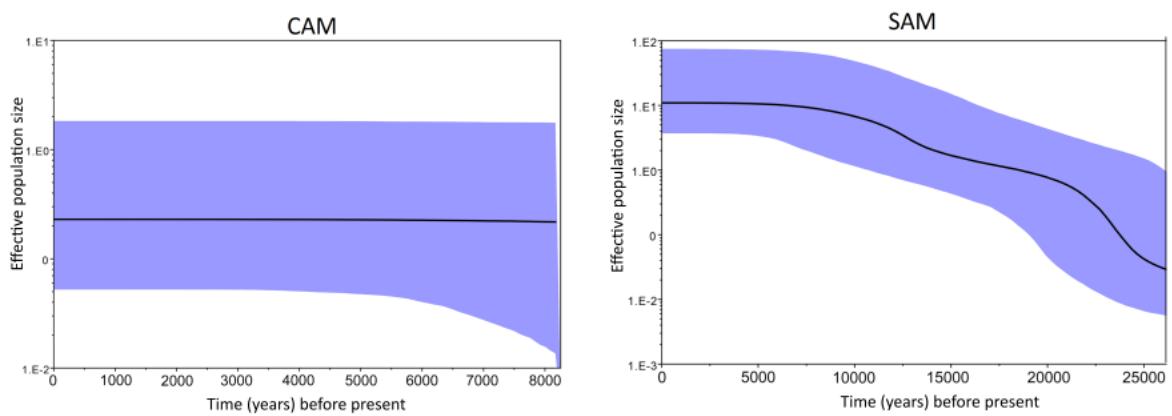
Supplementary Fig. 8. Bayesian skyline plot of the global population of *Plasmodium falciparum* showing changes in effective population size N_e (shown in log scale on y-axis) over time (x-axis). The black line shows the median ancestral population size, whereas the colored blue region shows the 95% highest probability density (HPD) interval for this estimate.



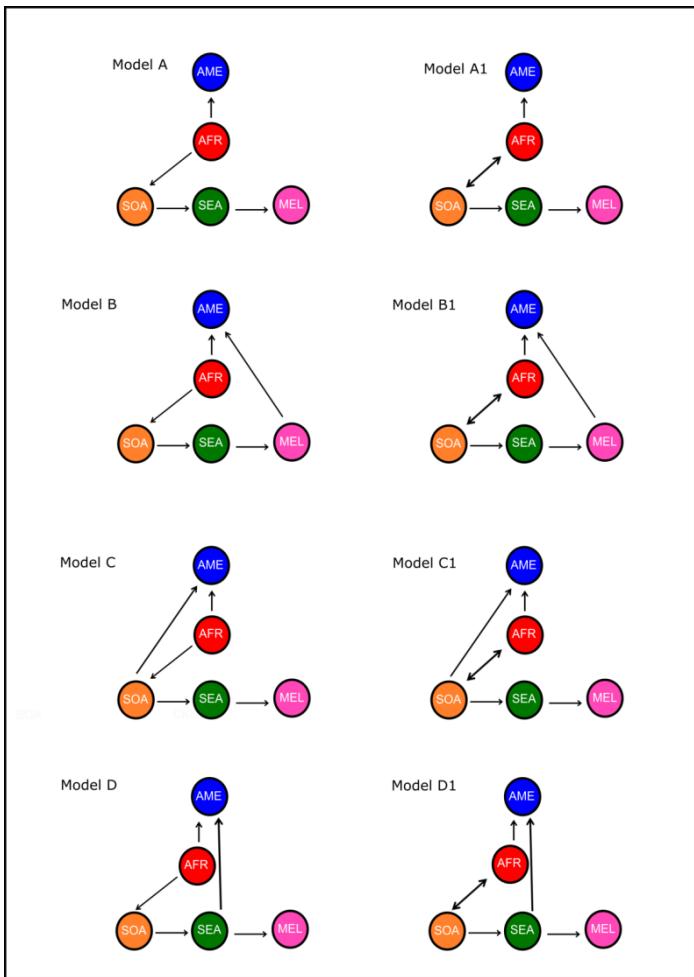
Supplementary Fig. 9. Bayesian skyline plot of the global population of *Plasmodium vivax* showing changes in effective population size N_e (shown in log scale on y-axis) over time (x-axis). The black line shows the median ancestral population size, whereas the colored blue region shows the 95% highest probability density (HPD) interval for this estimate.



Supplementary Fig. 10. Bayesian skyline plot of the SAM population of *Plasmodium falciparum* showing changes in effective population size N_e (shown in log scale on y-axis) over time (x-axis). The black line shows the median ancestral population size, whereas the colored blue region shows the 95% highest probability density (HPD) interval for this estimate.

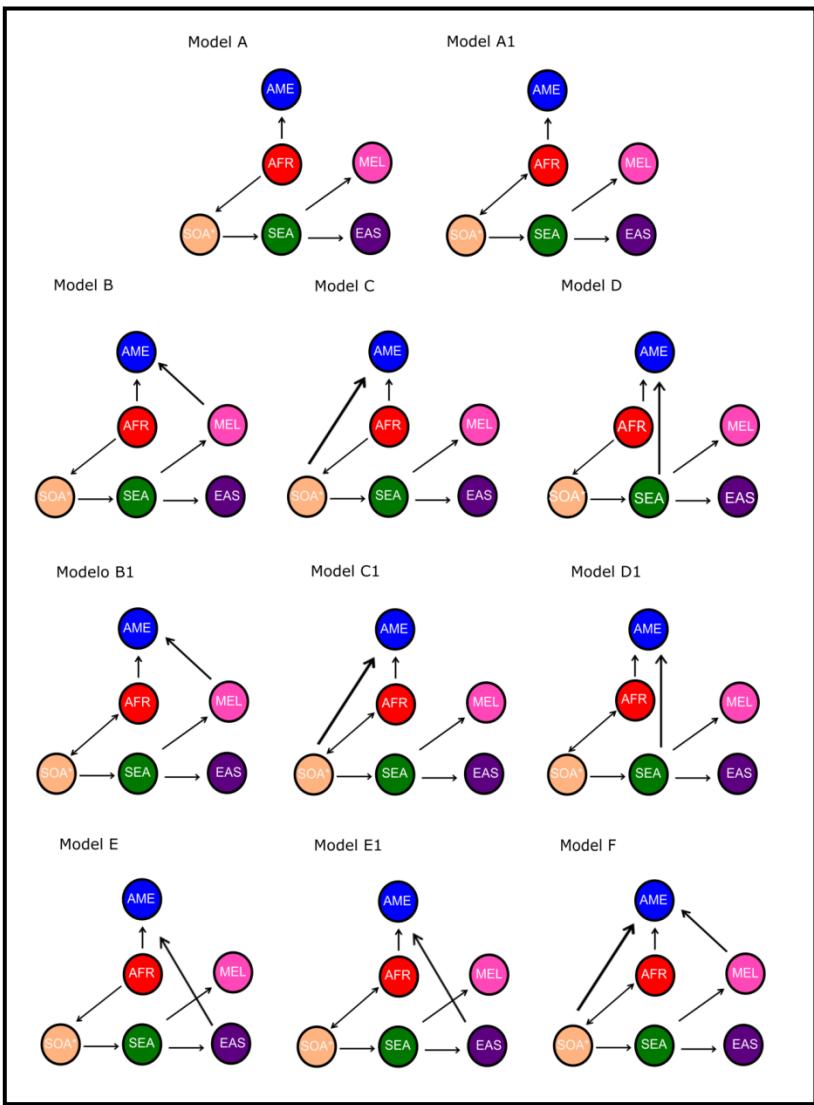


Supplementary Fig. 11. Bayesian skyline plot of the SAM and CAM populations of *Plasmodium vivax* showing changes in effective population size N_e (shown in log scale on y-axis) over time (x-axis). The black line shows the median ancestral population size, whereas the colored blue region shows the 95% highest probability density (HPD) interval for this estimate. SAM = South America; CAM = Central America and Mexico.

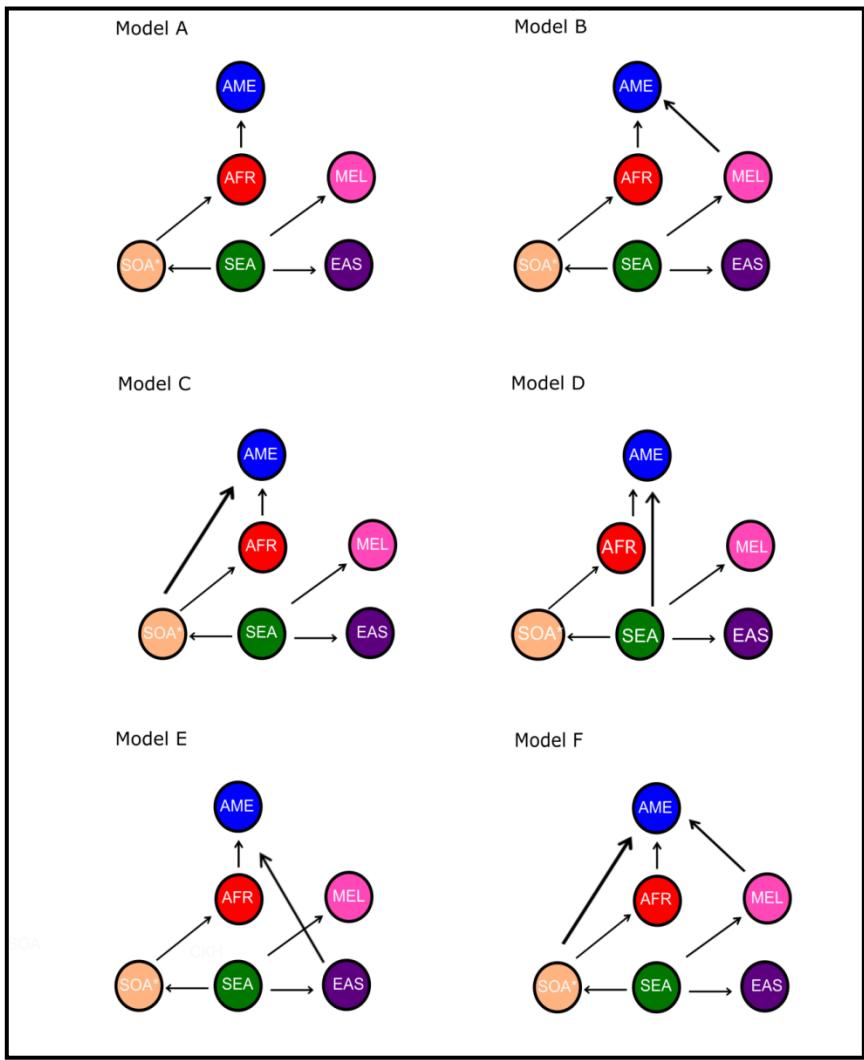


Supplementary Fig. 12. A priori *Plasmodium falciparum* migration models compared using Migrate-N to make inferences regarding gene flow between regional populations.

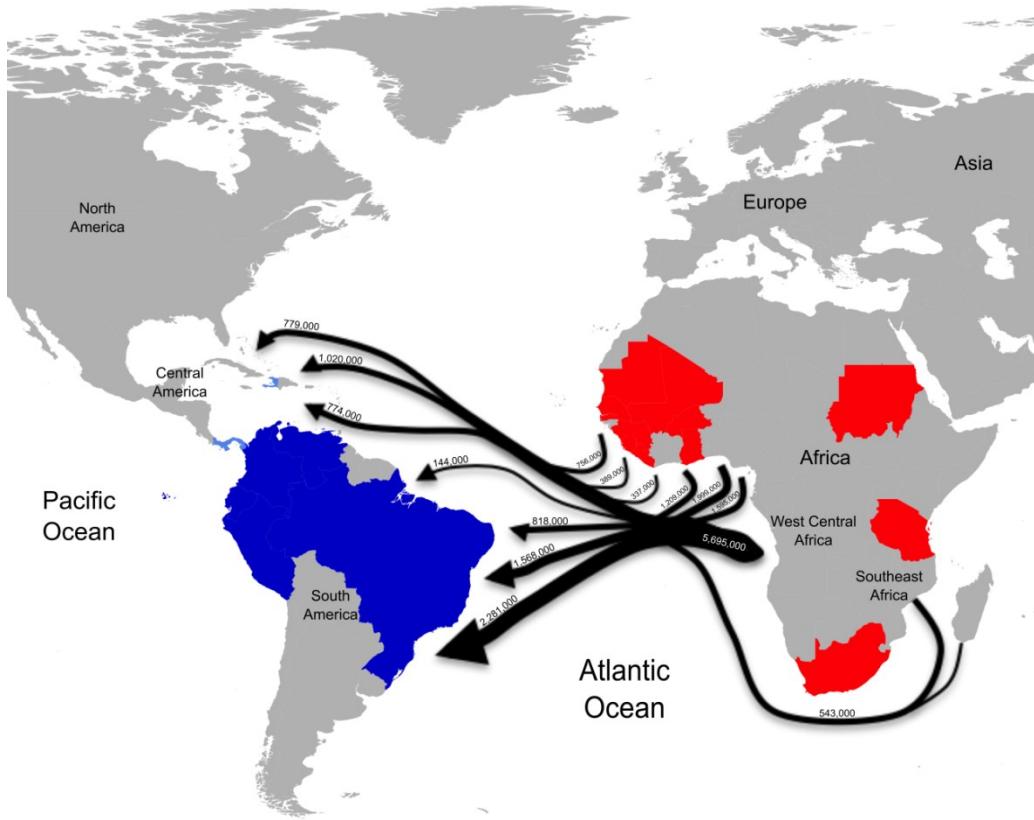
Colored circles represent the following regional populations: Africa (AFR; red), South and Central America combined (SAM; dark blue), South Asia (SOA; orange), Southeast Asia (SEA; green), and Melanesia (MEL; pink).



Supplementary Fig. 13. A priori migration models assuming an African origin of *Plasmodium vivax* that were compared using Migrate-N to make inferences regarding gene flow between regional populations. Colored circles represent the following populations: Africa (AFR; red), South and Central America (SAM; dark blue), Middle East, Central and South Asia combined (SOA*; light brown), Southeast Asia (SEA; green), East Asia (EAS; dark purple), and Melanesia (MEL; pink).



Supplementary Fig. 14. A priori migration models assuming a Southeast Asian origin of *Plasmodium vivax* that were compared using Migrate-N to make inferences regarding gene flow between regional populations. Colored circles represent the following populations: Africa (AFR; red), South and Central America (SAM; dark blue), Middle East, Central and South Asia combined (SOA*; light brown), Southeast Asia (SEA; green), East Asia (EAS; dark purple), and Melanesia (MEL; pink).



Supplementary Fig. 15. Map showing the geographic origins of enslaved Africans brought to the Americas between the 1500s and the mid-1800s. Figures next to arrows represent estimates of total numbers of slaves according to each route, which were derived from the Trans-Atlantic Slave Trade Database of Emory University (<http://www.slavevoyages.org>³⁵). The geographic origins of African and American mitochondrial lineages of *Plasmodium falciparum* analyzed in this study are indicated in the map at the country level using the same color code as that of Fig. 4a. Map built using the open-access R software library *rworldmap: mapping global data* combined with the *ggplot2* library, which are both available at <http://www.R-project.org/>.

Supplementary Tables

Supplementary Table 1. Country-specific levels of genetic diversity in *Plasmodium falciparum* mitogenomes from South America.

Country	No. isolates	Nucleotide diversity		H^d (SD) ^b
		π^a (SD) ^b	θs^c (SD) ^b	
Venezuela	21	0.00058 (0.00011)	0.00135 (0.00051)	0.900 (0.062)
Brazil	208	0.00014 (0.00002)	0.00079 (0.00022)	0.507 (0.041)
Total	229	0.00018 (0.00002)	0.00153 (0.00038)	0.559 (0.039)

^a π = average number of pairwise nucleotide differences per site; ^bSD = standard deviation; ^c θ_S

= standardized number of segregating sites; and ^d H = haplotype diversity.

Supplementary Table 2. Country-specific levels of genetic diversity in *Plasmodium vivax*

mitogenomes from South America.

Country	No. isolates	Nucleotide diversity		H^d (SD)
		π^a (SD) ^b	θ_S^c (SD) ^c	
Venezuela	15	0.00090 (0.00014)	0.00159 (0.00063)	1 (0.024)
Colombia	17	0.00066 (0.00015)	0.00148 (0.00058)	0.971 (0.032)
Peru	31	0.00058 (0.00006)	0.00151 (0.00052)	0.974 (0.020)
Brazil	171	0.00052 (0.00003)	0.00265 (0.00064)	0.904 (0.017)
Total	234	0.00058 (0.00003)	0.00328 (0.00075)	0.9323 (0.019)

^a π = average number of pairwise nucleotide differences per site; ^bSD = standard deviation; ^c θ_S = standardized number of segregating sites; and ^d H = haplotype diversity.

Supplementary Table 3. Number of *Plasmodium falciparum* mitogenome haplotypes that are unique within regional populations and shared between populations.

Regional population	Haplotypes			No. shared between populations					
	total	unique	shared	AFR ^a	SAM ^b	CAM ^c	SOA ^d	SEA ^e	MEL ^f
AFR ^a	191	179	12	-	4	2	5	7	5
SAM ^b	41	36	5		-	2	2	3	2
CAM ^c	5	2	3			-	1	1	1
SOA ^d	23	18	5			-		4	2
SEA ^e	56	48	8				-		4
MEL ^f	35	28	6					-	

^aAFR = Africa; ^bSAM = South America; ^cCAM = Central America; ^dSOA = South Asia;

^eSEA = Southeast Asia; and ^fMEL = Melanesia.

Supplementary Table 4. Pairwise genetic differentiation between mitogenomes of regional

Plasmodium falciparum populations as estimated by Wright's fixation index F_{ST} . All F_{ST}

values are significantly different from zero at the level of 0.05.

Population	AFR ^a	SAM ^b	CAM ^c	SOA ^d	SEA ^e	MEL ^f
AFR ^a	-					
SAM ^b	0.468	-				
CAM ^c	0.293	0.669	-			
SOA ^d	0.091	0.537	0.234	-		
SEA ^e	0.270	0.596	0.382	0.266	-	
MEL ^f	0.130	0.558	0.292	0.147	0.097	-

^aAFR = Africa; ^bSAM = South America; ^cCAM = Central America; ^dSOA = South Asia;

^eSEA = Southeast Asia; and ^fMEL = Melanesia.

Supplementary Table 5. Number of *Plasmodium vivax* mitogenome haplotypes that are unique within regional populations and shared between populations.

Population	Haplotypes			No. shared between populations							
	total	unique	shared	AFR ^a	SAM ^b	CAM ^c	SOA ^d	SEA ^e	MEL ^f	MCA ^g	EAS ^h
AFR ^a	45	39	6	-	0	2	5	0	1	1	0
SAM ^b	119	116	3	-	2	1	2	1	0	0	0
CAM ^c	12	7	5	-	2	1	1	1	2	0	0
SOA ^d	62	54	8	-	-	1	3	2	0	0	0
SEA ^e	87	80	7	-	-	-	4	0	2	0	0
MEL ^f	51	43	8	-	-	-	-	1	1	0	0
MCA ^g	19	16	3	-	-	-	-	-	0	0	0
EAS ^h	37	34	3	-	-	-	-	-	-	-	0

^aAFR = Africa; ^bSAM = South America; ^cCAM = Central America and Mexico; ^dSOA =

South Asia; ^eSEA = Southeast Asia; ^fMEL = Melanesia; ^gMCA = Middle East and Central

Asia; and ^hEAS = East Asia.

Supplementary Table 6. Pairwise genetic differentiation between mitogenomes of regional *Plasmodium vivax* populations, as estimated by Wright's fixation index F_{ST} . All F_{ST} values are significantly different from zero at the level of 0.05.

Population	AFR ^a	SAM ^b	CAM ^c	SOA ^d	SEA ^e	MEL ^f	MCA ^g	EAS ^h
AFR ^a	-							
SAM ^b	0.262	-						
CAM ^c	0.343	0.050	-					
SOA ^d	0.014	0.228	0.260	-				
SEA ^e	0.241	0.245	0.228	0.211	-			
MEL ^f	0.406	0.242	0.345	0.360	0.320	-		
MCA ^g	0.087	0.269	0.400	0.075	0.212	0.389	-	
EAS ^h	0.345	0.457	0.414	0.343	0.203	0.470	0.305	-

^aAFR = Africa; ^bSAM = South America; ^cCAM = Central America and Mexico; ^dSOA = South Asia; ^eSEA = Southeast Asia; ^fMEL = Melanesia; ^gMCA = Middle East and Central Asia; and ^hEAS = East Asia.

Supplementary Table 7. Place and date of collection and original host of 32 *P. vivax*/*P. simium* isolates from the Atlantic Forest of South and Southeast Brazil.

Isolate code	Place (year) of collection	Host	Haplotype	Reference
Fonseca MRA-353	Cantareira State Park, São Paulo (1960s)	Howler monkey (<i>Alouatta clamitans</i>)	AtI2	Jongwutiwes <i>et al.</i> ¹
SS3Ps	Cantareira State Park, São Paulo (2007)	Howler monkey (<i>Alouatta clamitans</i>)	AtI1	Duarte <i>et al.</i> ²
139Ps	Parelheiros, São Paulo (2003)	Howler monkey (<i>Alouatta clamitans</i>)	AtI1	Duarte <i>et al.</i> ²
BL65Ps	Indaial, Santa Catarina (2011)	Howler monkey (<i>Alouatta clamitans</i>)	AtI1	Costa <i>et al.</i> ³
RSPs	Itapuã State Park, Rio Grande do Sul (2003)	Howler monkey (<i>Alouatta clamitans</i>)	AtI1	Duarte <i>et al.</i> unpublished
160Ps	Cantareira State Park, São Paulo (2010)	Black-fronted titi monkey (<i>Callicebus nigrifrons</i>)	AtI1	Bueno ⁴
121Ps	Cantareira State Park, São Paulo (2003)	Howler monkey (<i>Alouatta clamitans</i>)	AtI1	Duarte <i>et al.</i> ²
95Ps	Cantareira State Park, São Paulo (2009)	Howler monkey (<i>Alouatta clamitans</i>)	AtI1	Yamasaki <i>et al.</i> ⁵
97Ps	Mariporã, São Paulo (2009)	Howler monkey (<i>Alouatta clamitans</i>)	AtI1	Yamasaki <i>et al.</i> ⁵
45Ps	Parelheiros, São Paulo (2007)	Howler monkey (<i>Alouatta clamitans</i>)	AtI1	Duarte <i>et al.</i> ²
1312MT	Santa Maria de Jetibá, Espírito Santo (2003)	Human	AtI1	Cerutti <i>et al.</i> ⁶
JSB62MT	Santa Teresa, Espírito Santo (2002)	Human	other	Cerutti <i>et al.</i> ⁶
1565MT	Santa Leopoldina, Espírito Santo (2003)	Human	AtI1	Cerutti <i>et al.</i> ⁶
GAB847MT	Santa Maria de Jetibá, Espírito Santo (2002)	Human	AtI2	Cerutti <i>et al.</i> ⁶
RO54MT	Domingos Martins, Espírito Santo (2002)	Human	other	Cerutti <i>et al.</i> ⁶
VC57MT	Santa Teresa, Espírito Santo (2002)	Human	AtI1	Cerutti <i>et al.</i> ⁶
1272MT	Alfredo Chaves, Espírito Santo (2003)	Human	AtI2	Cerutti <i>et al.</i> ⁶
OJA51MT	Santa Teresa, Espírito Santo (2002)	Human	AtI1	Cerutti <i>et al.</i> ⁶
1411MT	Santa Leopoldina, Espírito Santo (2003)	Human	AtI2	Cerutti <i>et al.</i> ⁶
1760MT	Espírito Santo	Human	AtI2	Cerutti <i>et al.</i> ⁶
1451MT	Santa Teresa, Espírito Santo (2003)	Human	AtI2	Cerutti <i>et al.</i> ⁶
ACC54MT	Santa Teresa, Espírito Santo (2002)	Human	AtI1	Cerutti <i>et al.</i> ⁶
FW63MT	Santa Teresa, Espírito Santo (2002)	Human	AtI2	Cerutti <i>et al.</i> ⁶
MA5M61MT	Espírito Santo	Human	other	Cerutti <i>et al.</i> ⁶
SV555MT	Marechal Floriano, Espírito Santo (2002)	Human	AtI1	Cerutti <i>et al.</i> ⁶
143MT	Santa Teresa, Espírito Santo (2002)	Human	AtI2	Cerutti <i>et al.</i> ⁶

40MT	Santa Teresa, Espírito Santo (2002)	Human	<i>Ame1</i>	Cerutti <i>et al.</i> ⁶
103_03MT	Espírito Santo	Human	<i>Ame1</i>	Cerutti <i>et al.</i> ⁶
111MT	Santa Teresa, Espírito Santo (2002)	Human	<i>Atl2</i>	Cerutti <i>et al.</i> ⁶
761MT	Santa Leopoldina, Espírito Santo (2002)	Human	other	Cerutti <i>et al.</i> ⁶
ALNL53MT	Santa Teresa, Espírito Santo (2002)	Human	other	Cerutti <i>et al.</i> ⁶
AJ54MT	Santa Teresa, Espírito Santo (2002)	Human	<i>Atl2</i>	Cerutti <i>et al.</i> ⁶

Supplementary Table 8. Private single-nucleotide polymorphisms (indicated by boldface letters) in the mitogenome of 32 *P. vivax*/*P. simium* isolates from the Atlantic Forest of South and Southeast Brazil.

Isolate code	Nucleotide position (gene)				
	C1342G	A3325T	T4134C	A4468G	A5322C
	Intergenic	<i>cox1</i>	<i>cox1</i>	<i>cox1</i>	<i>cytb</i>
Fonseca MRA-353	C	T	C	G	A
SS3Ps	C	A	C	G	A
139Ps	C	A	C	G	A
BI65Ps	C	A	C	G	A
R5Ps	C	A	C	G	A
160Ps	C	A	C	G	A
121Ps	C	A	C	G	A
95Ps	C	A	C	G	A
97Ps	C	A	C	G	A
45Ps	C	A	C	G	A
1312MT	C	A	C	G	A
JSB62_MT	G	A	C	G	A
1565MT	C	A	C	G	A
GAB847_MT	C	T	C	G	A
RO54_MT	G	A	C	G	A
VC57MT	C	A	C	G	A
1272MT	C	T	C	G	A
OJA51_MT	C	A	C	G	A
1411MT	C	T	C	G	A
1760MT	C	T	C	G	A
1451MT	C	T	C	G	A
ACC54_MT	C	A	C	G	A
FW63MT	C	T	C	G	A
MA5M61_MT	C	T	T	G	A
SV555_MT	C	A	C	G	A
143MT	C	T	C	G	A
40MT	C	A	T	A	A
103_03MT	C	A	T	A	A
111MT	C	T	C	G	A
761MT	C	T	C	A	A
ALNL53MT	C	T	T	G	C
AJR54_MT	C	T	C	G	A

Supplementary Table 9. The results of Tajima's D and Fu's F_s neutrality tests applied to *Plasmodium falciparum* mitogenomes from South America. Statistically significant P values are underlined.

Country	Neutrality test			
	Tajima's D	<i>P value</i>	Fu's F_s	<i>P value</i>
Venezuela	-2.18761	<u>0.006</u>	-3.2007	<u><0.02</u>
Brazil	-2.34520	<u><0.0001</u>	-5.05091	<u><0.02</u>
Total	-2.61249	<u>0.001</u>	-8.47147	<u><0.02</u>

Supplementary Table 10. The results of Tajima's D and Fu's F_s neutrality tests applied to *Plasmodium vivax* mitogenomes from South America. Statistically significant P values are underlined.

Country	Neutrality test			
	Tajima's D	P value	Fu's F_s	P value
Venezuela	-1.84870	<u>0.01</u>	-2.46987	<0.05
Colombia	-2.26611	<u><0.0001</u>	-3.19186	<u><0.02</u>
Peru	-4.76892	<u><0.0001</u>	-3.90115	<u><0.02</u>
Brazil	-2.50856	<u><0.0001</u>	-6.25391	<u><0.02</u>
Total	-2.52771	<u>0.001</u>	0.16976	>0.10

Supplementary Table 11. Sum of square deviations (SDD) and raggedness index (R) comparing the observed mismatch distribution in each of the regional populations of *Plasmodium falciparum* and *P. vivax* with the expected distribution under a sudden demographic expansion model; *P* values > 0.05 indicate an expanding population.

Population	<i>P. falciparum</i>		<i>P. vivax</i>	
	SSD (<i>P</i> value)	R (<i>P</i> value)	SSD (<i>P</i> value)	R (<i>P</i> value)
AFR ^a	0.00267 (<0.001)	0.07110 (<0.001)	0.00237 (0.88)	0.01068 (0.97)
SAM ^b	0.00043 (0.80)	0.05627 (0.81)	0.00024 (0.95)	0.01488 (0.80)
EAS ^c	-	-	0.00583 (0.64)	0.00866 (0.89)
MCA ^d	-	-	0.00357 (0.43)	0.02112 (0.52)
MEL ^e	0.00916 (0.16)	0.05466 (0.26)	0.00124 (0.68)	0.02483 (0.43)
CAM ^f	0.01577 (0.44)	0.08163 (0.56)	0.00023 (0.52)	0.17341 (0.79)
SEA ^g	0.01135 (0.22)	0.06169 (0.28)	0.00594 (0.17)	0.00852 (0.75)
SOA ^h	0.00287 (0.12)	0.04055 (0.03)	0.00023 (0.78)	0.02336 (0.38)

^aAFR = Africa; ^bSAM = South America; ^cEAS = East Asia (only *P. vivax*); ^dMCA = Middle East and Central Asia (only *P. vivax*); ^eMEL = Melanesia; ^fCAM = Central America and Mexico; ^gSEA = Southeast Asia; and ^hSOA = South Asia.

Supplementary Table 12. Sum of square deviations (SDD) and raggedness index (R) comparing the observed mismatch distribution for South American populations of *Plasmodium falciparum* and *P. vivax* with the expected distribution under a sudden demographic expansion model; *P* values > 0.05 indicate an expanding population.

Country	<i>P. falciparum</i>		<i>P. vivax</i>	
	SSD (<i>P</i> value)	R (<i>P</i> value)	SSD (<i>P</i> value)	R (<i>P</i> value)
Brazil	0.00005 (0.88)	0.07416 (0.76)	0.00013 (1.00)	0.01671 (0.87)
Colombia	-	-	0.00894 (0.45)	0.03492 (0.43)
Peru	-	-	0.00362 (0.09)	0.02729 (0.26)
Venezuela	0.01168 (0.58)	0.02066 (0.92)	-	-

Supplementary Table 13. Comparison of *Plasmodium falciparum* migration models

estimated by Migrate-N; models are described in Supplementary Fig. 12.

Model	No. parameters ^a	Bézier log mL ^b	LBF ^c	Model probability
A	4 M , 5 Θ	-14736.87	-93.19	<0.00000001
B	5 M , 5 Θ	-14782.59	-138.91	<0.00000001
C	5 M , 5 Θ	-14732.10	-88.42	<0.00000001
D	5 M , 5 Θ	-14740.11	-96.43	<0.00000001
A1	5 M , 5 Θ	-14644.82	-1.14	0.24030838
B1	6 M , 5 Θ	-14648.19	-4.51	0.00825825
C1	6 M , 5 Θ	-14643.68	0.00	0.75143332
D1	6 M , 5 Θ	-14660.09	-16.41	0.00000005

^aNumber of parameters estimated in the model: M = median mutation-scaled pairwisemigration rate and Θ = median mutation-scaled effective population size; ^bBézier log mL =Bézier approximation of log marginal likelihood; ^cLBF = log Bayes factor.

Supplementary Table 14. Comparison of migration models assuming an African origin of *Plasmodium vivax* estimated with Migrate-N; models are described in Supplementary Fig. 13.

Model	No. parameters ^a	Bézier log mL ^b	LBF ^c	Model probability
A	5 M , 6 Θ	-15984.80	-53.92	<0.0000001
B	6 M , 6 Θ	-15958.54	-27.66	<0.0000001
C	6 M , 6 Θ	-15969.08	-38.20	<0.0000001
D	6 M , 6 Θ	-15980.13	-49.25	<0.0000001
E	6 M , 6 Θ	-15957.98	-27.10	<0.0000001
A1	6 M , 6 Θ	-15933.73	-2.850	0.0533535
B1	7 M , 6 Θ	-15934.53	-3.65	0.0239896
C1	7 M , 6 Θ	-15938.62	-7.74	0.0003817
D1	7 M , 6 Θ	-16001.45	-70.57	<0.0000001
E1	7 M , 6 Θ	-15964.90	-34.02	<0.0000001
F	8 M , 6 Θ	-15930.88	0.00	0.9223650

^aNumber of parameters estimated in the model; M = median mutation-scaled pairwise

migration rate and Θ = median mutation-scaled effective population size; ^bBézier log mL =

Bézier approximation of log marginal likelihood; ^cLBF = log Bayes factor.

Supplementary Table 15. Comparison of migration models assuming a Southeast Asian origin of *Plasmodium vivax* estimated with Migrate-N; models are described in Supplementary Fig. 14.

Model	No. parameters ^a	Bézier log mL ^b	LBF ^c	Model probability
A	5 M, 6 Θ	-15929,99811	-16.64	<0.00000001
B	6 M, 6 Θ	-15913,36034	0.00	0.99999500
C	6 M, 6 Θ	-15969,51376	-56.15	<0.00000001
D	6 M, 6 Θ	-15963,05640	-49.70	<0.00000001
E	6 M, 6 Θ	-15936,12640	-22.77	<0.00000001
F	7 M, 6 Θ	-15925,55080	-12.19	0.00000499

^aNumber of parameters estimated in the model: M = median mutation-scaled pairwise migration rate and Θ = median mutation-scaled effective population size; ^bBézier log mL = Bézier approximation of log marginal likelihood; ^cLBF = log Bayes factor.

Supplementary Table 16. Oligonucleotide primers used to amplify and sequence the mitogenomes.

<i>P. falciparum - amplification and sequencing</i>		
Primer name	Sequence (5' - 3')	Amplicon size
PfmtDNA1amp	F:5'-CGGGCAGATGTCAGTAACCT-3' R:5'-AGGGCTTAAACCAACAAACAT-3'	2001 bp
PfmtDNA2amp	F:5'-AGAACTCCAGGCAGTTAACGT-3' R:5'-CACCTACCCTCCATAATTCTC-3'	2013 bp
PfmtDNA3amp	F:5'-CAGGTGTATTTTAGCAAGTCG-3' R:5'-GGCGCTTCCATTATAAGAA-3'	2268 bp
PfmtDNA1seq	F:5'-CGGGCAGATGTCAGTAACCT-3' R:5'-TGAGCTGGGTTAAGAACGTC-3'	505 bp
PfmtDNA2seq	F:5'-CAAACCTGCCCTCAAGACGTT-3' R:5'-TTGGCATTGTTGAAATAGTC-3'	490 bp
PfmtDNA3seq	F:5'-TTTGGCATTGTTGAAATAGTC-3' R:5'-TTATTCTTACATATTTTGGGTAT-3'	495 bp
PfmtDNA4seq	F:5'-CAGAATAAAAACTTCTCGAATAGAA-3' R:5'-AGGGCTTAAACCAACAAACAT-3'	516 bp
PfmtDNA5seq	F:5'-AGAACTCCAGGCAGTTAACCT-3' R:5'-CATTTCTTGTGCAATTATTCTT-3'	510 bp
PfmtDNA6seq	F:5'-ACGTACTGAATTATATTCTTCATCTT-3' R:5'-TGTTAGCAATAACATTCTGATG-3'	496 bp
PfmtDNA7seq	F:5'-TTATTGCTAACACTACCGGTTT-3' R:5'-ACATCAATGGCAGCATTACC-3'	512 bp
PfmtDNA8seq	F:5'-GGTAATGCTGCCATTGATGT-3' R:5'-TGCATATGAAACATCTGGTG-3'	530 bp
PfmtDNA9seq	F:5'-CAGGTGTATTTTAGCAAGTCG-3' R:5'-CCCTAAAGGATTGTGCTACC-3'	479 bp
PfmtDNA10seq	F:5'-TTCTTACATTTACATGGTAGCACAA-3' R:5'-ATCTTGTGGAATTGACATCCA-3'	450 bp
PfmtDNA11 seq	F:5'-TTGGATGTCATTACCAACAGA-3' R:5'-GCATGCAATACCGAACATTT-3'	495 bp
PfmtDNA12seq	F:5'-GTATTGCATGCCCTGGTGT-3' R:5'-AAAAATGGCTGCTGGAAAGTA-3'	482 bp
PfmtDNA13seq	F:5'-TTCTACTCCAGCAGCCATT-3' R:5'-ATGGAGCACTGGATTGGATA-3'	486 bp
<i>P. vivax - amplification and sequencing</i>		
Primer	Sequence (5' - 3')	Size
Pv mtDNA1amp	F:5'-CGCTGACTTCCTGGCTAC-3' R:5'-GTCAGGCAGTTAAAGCGTTC-3'	2911 bp
Pv mtDNA2amp	F:5'-TTGTACACACCGCTCGTCAC-3' R:5'-CCGAAACCTGGACTCTGAA-3'	3497 bp
Pv mtDNA1seq	F:5'ACGCTGACTTCCTGGCTAA-3' R:5'GGATGAAACCTTCCTGATCG-3'	540 bp
Pv mtDNA2seq	F:5'CACGAGTCGATCAGGAAGGT-3' R:5'GTGTTGGCTGGCATTAGTC-3'	536 bp
Pv mtDNA3seq	F:5'GACTAATGCCAGCCAACAC-3' R:5'GCTATCAAATGGCGAGAAGG-3'	566 bp

Pv mtDNA4seq	F:5'CCTTCTGCCATTGATAGC-3' R:5'GCATCATGTATGACAGCATGTT-3'	524 bp
Pv mtDNA5seq	F:5'TGCTGTCATACATGATGCACTT-3' R:5'CAAGGCAACAATAACACGCTAA-3'	554 bp
Pv mtDNA6seq	F:5'AGCGTGTATTGTTGCCTTGT-3' R:5'CATCCATGTCAGGCGTTAAA-3'	303 bp
Pv mtDNA7seq	F:5'TTGTACACACCGCTCGTCAC-3' R:5'AACTACCAAATAAAATGAAAACCA-3'	495 bp
Pv mtDNA8seq	F:5'CAAATTGCAATCATAAAACTTAGGTC-3' R:5'CTAGCAATACCAGATACTAAAAGACCA-3'	460 bp
Pv mtDNA9seq	F:5'TCATTGTTGGTCTTTAGTATCTGG-3' R:5'CCAATTAAATATTTTGTTCAGTAGG-3'	530 bp
Pv mtDNA10seq	F:5'CCTACTGGAACAAAATATTAATTGG-3' R:5'TTAATGGGCATGGTAATT-3'	557 bp
Pv mtDNA11seq	F:5'AAATTACCCATGCCATTAAA-3' R:5'CCCTAAAGGATTGTGCTACC-3'	562 bp
Pv mtDNA12seq	F:5'TGGTAGCACAAATCCTTAGGG-3' R:5'AAATGTTGCTTGGGAGCTG-3'	548 bp
Pv mtDNA13seq	F:5'ACAGCTCCCAGAACACATT-3' R:5'GACCGAACCTTGGACTCTG-3'	539 bp

P. simium - amplification and sequencing

Primer	Sequence (5' - 3')	Size
Amp1Ps	F:5'-TCCACACTCAATTGACTTCC-3' R:5'-GCAAACACTAGCGGTGGAAT-3'	1147 bp
Nest1_1Ps	F:5'-ACAAAATATAATCTCCTGTTCTAATG-3' R:5'-CAAGTCACTGATAATTCCGATGA-3'	533 bp
Nest1_2Ps	F:5'-CGGGCAGATGTCAGTAACTTG-3' R:5'-TTGTTTCATTGATAGTAAACGCTAT-3'	635 bp
Amp2Ps	F:5'-GCTCACGCATCGCTTCTAAC-3' R:5'-GATGAGACGACATGGAGGTG-3'	1287 bp
Nest2_1Ps	F:5'-TGGAACCTGTTCAAGTTCAAAT-3' R:5'-TGCATCACTCAAACAACGTGAA-3'	665 bp
Nest2_2Ps	F:5'-CCTGTAACACAATAAAATAATGTACCA-3' R:5'-CCCGCGAACCTTCTTAC-3'	602 bp
Amp3Ps	F:5'-GCCGGGGATAACAGGTTATAGT-3' R:5'-TCTCCAGCAAATGTAGGATCAA-3'	1221 bp
Nest3_1Ps	F:5'-GCCGGGGATAACAGGTTATAGT-3' R:5'-CCTAAAGTTTATGATTGCAATTGT-3'	588 bp
Nest3_2Ps	F:5'-CAAATTGCAATCATAAAACTTGTAGTC-3' R:5'-TGATAATAACATTAAAACACCACCTG-3'	623 bp
Amp4Ps	F:5'-TGCATCTAACAGATCTAACAGGTTAAC-3' R:5'-TTTGTCCCCAAGGTAAAACG-3'	1326 bp
Nest4_1Ps	F:5'-TGTTTAACAGGTGGTGTGTTAATG-3' R:5'-GAATTTCACGTAATGTTACCAA-3'	627 bp
Nest4_2Ps	F:5'-AGGTGCAATTATTGGATTGTTA-3' R:5'-AAGGTAAAACGTAACCTATAAAAGCTG-3'	674 bp
Amp5Ps	F:5'-TGCTACAGGTGCATCTCTGTATT-3' R:5'-CAAAATGGCTGCTGGAAAGTA-3'	1636 bp
Nest5_1Ps	F:5'-CGTTTACCTTGGGGACAAA-3' R:5'-TGAACAAGTGCAAATAATCCAC-3'	708 bp

Nest5_2Ps

F:5'-TTGGATGTCAGTTACCACAAGA-3'

765 bp

R:5'-ACGCGAAAAACCCCTAGAACCA-3'

Supplementary Text 1: 18S rRNA gene sequence types in *Plasmodium vivax* isolates from the Amazon Basin of Brazil (Rondônia and Acre) and Sri Lanka (Kataragama)

Li et al. (7) have suggested that a single gene conversion event in the gene coding for the S-type of the 18S rRNA can be used to distinguish Old World (wild-type) from New World (variant) lineages of *P. vivax*. However, unpublished sequence data deposited in the GenBank database in 2005 by F. E. Merino and H. A. del Portillo (then at the University of São Paulo, Brazil) show that both Old World and New World types of 18S rRNA gene sequences are found in the Amazon Basin of Brazil (Rondônia and Acre) and in Sri Lanka (Kataragama), thus indicating that neither variant sequences are limited to the New World nor wild-type sequences circulate exclusively in the Old World. These data are summarized below.

GenBank accession	Sample code	Country: site	18S sequence type
DQ162614	MalDB36	Brazil: Rondonia	New World
DQ162600	MalDB37	Brazil: Rondonia	New World
DQ162604	MalDB39	Brazil: Rondonia	New World
DQ162598	MalDB56	Brazil: Rondonia	New World
DQ162608	MalDB58	Brazil: Rondonia	New World
DQ162615	MalDB59	Brazil: Rondonia	New World
DQ162602	MalDB60	Brazil: Rondonia	New World
DQ162599	MalDB63	Brazil: Rondonia	New World
DQ162603	MalDB65	Brazil: Rondonia	New World
DQ162601	MalDB66	Brazil: Rondonia	New World
DQ162611	MalDB176	Brazil: Acre	New World
DQ162607	MalDB177	Brazil: Acre	New World
DQ162612	MalDB178	Brazil: Acre	New World
DQ162609	MalDB179	Brazil: Acre	New World
DQ162613	MalDB180	Brazil: Acre	New World
DQ162595	MalDB181	Brazil: Acre	New World
DQ162610	MalDB182	Brazil: Acre	New World
DQ162606	MalDB184	Sri Lanka: Kataragama	New World
DQ162596	MalDB185	Sri Lanka: Kataragama	New World
DQ162597	MalDB187	Sri Lanka: Kataragama	New World
DQ162605	MalDB186	Brazil: Acre	New World
DQ162619	MalDB183	Sri Lanka: Kataragama	Old World

DQ162620	MalDB54	Brazil: Rondonia	Old World
DQ162616	MalDB41	Brazil: Rondonia	Old World
DQ162617	MalDB57	Brazil: Rondonia	Old World
DQ162618	MalDB61	Brazil: Rondonia	Old World

Supplementary References

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